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## Title Page

### **Dentinal tubules occluded by bioactive-glass containing toothpaste exhibit high resistance toward acidic soft drink challenge**

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## **Abstract**

*Background:* Dentine hypersensitivity is a common problem attributed by patent dentinal tubules. Ingredients incorporated in toothpastes aim to occlude patent dentinal tubules to minimise the dentine hypersensitivity. However, frequent consumption of acidic soft drinks may reverse the dentinal tubules occlusion. In this *in vitro* study, the efficacy of dentinal tubules occluded by commercially available toothpastes to withstand different duration of an acidic soft drink challenge was investigated.

*Methods:* One hundred and twenty dentine discs were divided into three groups. The discs from each group were brushed with toothpaste containing bioactive glass, arginine and control toothpaste. Each group were then divided into four subgroups and exposed to acidic soft drink over four different time durations.

*Results:* The scoring and the percentage of occluded dentinal tubules by Novamin containing toothpaste was significantly better compared to arginine or the control toothpaste. Acidic soft drink challenge reduced the extent of dentinal tubules occlusion along with time. Dentinal tubules occluded by Novamin containing toothpaste withstand the acidic challenge comparatively for longer period.

*Conclusions:* The findings demonstrated that occlusion of dentinal tubules is more efficient by the bioactive-glass containing toothpaste and thus may contribute to its better resistance to acidic soft drink challenge.

**Key words:** Acidic soft drink challenge; arginine containing toothpaste; bioactive-glass (Novamin) containing toothpaste; dentine hypersensitivity; dentinal tubule.

## **Introduction**

Dentin hypersensitivity is a common oral health problem which can affect many patients. It is characterized by a short and sharp pain arising from the tooth with exposed dentine in response to stimuli such as thermal, evaporative, tactile, osmotic or chemical.<sup>1,2</sup> The dentine may be exposed as a result of attrition, erosion, abrasion, gingival recession or periodontal disease.<sup>2</sup> It has been reported that in hypersensitive teeth, a large proportion of the dentinal tubules has become patent thus, allowing direct contact from the oral environment to the pulp.<sup>1-4</sup> The most acceptable theory for the mechanism of dentine hypersensitivity, proposed by Brannstrom and Astrom<sup>5</sup>, states that dentine hypersensitivity occurs due to movement of the fluid within the dentinal tubules which could be triggered by an external stimuli, which in turn stimulates the nerves of the pulp (hydrodynamic theory). In order to reduce fluid movement, one strategy that has been used to manage dentine hypersensitivity is to occlude patent dentinal tubules.<sup>6</sup> Active ingredients such as Novamin<sup>7-11</sup> and arginine<sup>12-14</sup> are being incorporated in commercially available toothpastes to create smear layers or smear plugs to occlude these patent tubules thereby reducing dentine hypersensitivity.<sup>6</sup> The former made up of bioactive glass as the active ingredient and was originally developed for regeneration of bone and contains calcium, sodium, phosphate and silica as its amorphous matrix.<sup>7</sup> Novamin based toothpastes has been clinically shown to reduce dentine hypersensitivity.<sup>8-11</sup> Arginine based toothpastes contains arginine and calcium carbonate as the active ingredients and has been reported to be able to occlude patent dentinal tubules.<sup>12-14</sup>

Although the use of dentifrices can occlude the dentinal tubules, exposure to acidic beverages consumed in daily life could reverse or decrease tubule occlusion by removing the smear layer.<sup>15-18</sup> In the present modern lifestyle, there is an increase in the consumption of acidic beverages, where frequent exposure of teeth to acid challenges has been observed to increase the incidence of dentine hypersensitivity.<sup>3,4</sup> It is thus worthy to search for active ingredients

that are able to not only occlude the dentinal tubules but also withstand acid challenge. An *in vitro* study conducted using dentine discs of bovine incisor teeth<sup>19</sup> showed that Novamin containing toothpastes impart significant tubule occlusion. It was also found that these positive effects were significantly better than the arginine based toothpaste.<sup>19</sup> Nevertheless, it has also been reported that there were no significant difference between the performances of these two toothpastes in withstanding acid challenge.<sup>20</sup> In order to have a better understanding of the acid withstanding efficacy of the occluded dentinal tubules brushed by commercially available toothpastes, this *in vitro* study was designed to assess the occluding efficacy of Novamin and arginine containing toothpastes and to evaluate the resistance capacity of the occluded tubules to an acidic soft drink challenge at various time point of exposure.

## **Materials and Methods:**

### **Preparation of dentine discs**

Dentine discs were prepared from freshly extracted human molars collected from the Oral Surgery Department, University Malaya, after obtaining informed consent from the patients. The protocol received approval by the Medical Ethics Committee, Faculty of Dentistry, University of Malaya. 120 dentine discs of 2 mm thickness were prepared by sectioning the teeth horizontally at the cervical area using a water-cooled diamond saw. The discs were sterilised in sodium hypochlorite (20,000 ppm) and washed in distilled water. The surfaces of each dentine disc were then smoothen using 600-grit silicon carbide paper to create flat surface before they were etched using 0.5 M EDTA solution (pH7.4) for 2 min to obtain patent dentine tubules. The discs were ultra-sonicated for 30 mins in distilled water and ensured that all dentinal tubules were fully patent. The discs were examined by SEM (Phenom G2 pro desktop SEM, Lambdaphoto, UK /Phenom-World).

## Experiment on tubule occlusion and acid challenge

The dentine discs were randomly divided into three groups of 40 discs. The discs were brushed for 2 mins with the different toothpastes. Discs in Group-1 were brushed with a commercially available Novamin containing; discs of Group-2 were brushed with a commercially available arginine containing toothpaste and discs of Group-3 were brushed with a control toothpaste which was a non-Novamin and non-arginine containing tooth paste. Compositions of the toothpastes used in this study are summarised in Table 1. A soft tooth brush (Oral B Power Toothbrush), and approximately 1g of the respective toothpastes at neat concentration (undiluted) was used in this process. The toothpaste-treated discs in each Group (1, 2 and 3) were further divided into four subgroups, each comprising of ten dentine discs. Discs in each subgroup were then exposed to acidic soft drink (Coca-Cola; pH 2.5) over four time durations of 0.5, 1, 2 and 5 mins.

## Evaluation of tubule occlusion and acid resistant

Each treated dentine discs were examined under the SEM by an examiner who was blinded to the study. Tubules occlusion was determined using a visual scoring index of 1 to 5, which was adopted from previous researches<sup>16-18</sup> (1-occluded; 2-partially unoccluded; 3-equally occluded/unoccluded; 4-partially occluded; 5-unoccluded) (Fig. 1). The percentage of occluded tubules (partially or completely covered with dentifrices) was also determined by using the following formula:

$$\frac{\text{No. of partially/completely covered dentinal tubules after treatment}}{\text{No. of patent dentinal tubules before any treatment}} \times 100$$

To determine the reliability of the scoring and counting of occluded dentinal tubules, forty dentine discs were randomly chosen and re-examined by the same examiner in addition to

other examiner. Intra- and inter-examiner reliability was determined using intra-class correlation coefficients. Intra-examiner and inter-examiner reliability coefficients for counting dentinal tubule occlusion were found to be substantial (0.87 and 0.79, respectively).

### **Statistical analysis**

For each of the toothpaste used in this study one way ANOVA followed by Dunnett's test were performed to compare the data obtained before and after the acidic soft drink challenge at different time points of soft drink challenge. Comparison of data among the three toothpastes was performed by one way ANOVA followed by Tukey Test. A p-value of less than 0.05 was considered statistically significant.

### **Results**

#### **Dentinal tubule occlusion by Novamin, arginine and control toothpastes**

Following treatment of dentine discs with various toothpastes, the highest number of occluded dentinal tubules was obtained with Novamin followed by the arginine and control toothpastes (Fig. 2, Fig. 3). The mean tubule occlusion score for Novamin was significantly lower (indicating better occlusion) than the other two toothpastes ( $p < 0.05$ ) (Fig. 3A, Table 2). Comparative to the control, the score for arginine containing toothpaste was lower but not statistically significant. The percentage of dentinal tubules covered with dentifrices (partially or completely) was also significantly higher for Novamin containing toothpaste compared to the control (Fig. 3B). However, the percentage difference between arginine and Novamin, and between arginine and control was not statistically significant.

#### **Dentinal tubule occlusion after exposure to acidic soft drink challenge**

For all types of toothpastes, the mean tubule occlusion score displayed gradual increment following acidic soft drink challenge (Fig. 3A, Table 2). After 0.5 min and subsequent exposure (1, 2, and 5 mins) to acidic soft drink, the tubule occlusion score was observed significantly different compared to the pre-exposure data for arginine containing and the control toothpastes. For the Novamin treated group however, no significant difference in the occlusion score was observed following the 0.5 min exposure, but significantly different scores were obtained following 1 min and subsequent exposure periods to the soft drink (Fig. 3A, Table 2). The percentage of occluded dentinal tubules for arginine and control toothpastes was also observed to be significantly lower after 0.5 mins and subsequent exposure of acidic soft drink challenge compared to pre-exposure data (Fig. 3B). In contrast for Novamin, the percentage significantly decreased after 2 mins and subsequent exposure to the challenge (Fig. 3B).

The dentinal tubule occlusion score among the three different types of toothpastes at different time points following acidic soft drink exposure showed that the score was significantly lower (indicating better occlusion) for Novamin compared to arginine containing and the control toothpastes at all time points (Fig. 3A, Table 2). However, the score was not significantly different between the control and arginine toothpastes. The percentage of occlusion was also significantly high for Novamin compared to arginine containing and the control toothpastes at all time points following acid challenge (Fig. 3B). The percentage of occlusion for arginine containing toothpaste was not significantly high compared to the control toothpastes except at the 1 min time point.

## Discussion

The present study shows that Novamin and arginine containing toothpastes and the control toothpastes can occlude considerable amount of patent dentinal tubules after two minutes of application. Upon comparison, Novamin containing toothpaste however, showed better dentinal tubule occluding efficacy compared to arginine containing and the control toothpastes. Dentinal tubules occluded by Novamin containing toothpaste also exhibited better resistance to acidic soft drink challenge.

Several *in vitro* and *in vivo* investigations have shown the efficacy of different toothpastes in occluding dentinal tubules.<sup>8-15</sup> The results obtained for the dentinal tubule occluding ability of both Novamin and arginine containing toothpastes are in accordance with previous studies. The amorphous sodium calcium phosphosilicate present in Novamin containing toothpaste showed a strong attraction for collagen.<sup>7, 8</sup> Due to the high collagen content of dentine Novamin binds to exposed dentine surfaces and physically occlude the dentine tubules.<sup>8</sup> Arginine on the other hand combines with calcium carbonate to form alkaline agglomerate which has high affinity for exposed dentinal surface and hence, resulting in occlusion of dentinal tubules.<sup>12</sup> Abrasive components in toothpastes may also play a role in occluding dentinal tubules.<sup>21-23</sup> Better occluding effect of Novamin than arginine has been reported earlier on bovine dentine discs.<sup>19</sup> Similar finding was observed in our study whereby brushing with Novamin containing toothpaste for 2 minutes gave a significantly better occluding score than the arginine containing tooth paste (Fig. 3A, Table 2). In contrast to our findings, an *in situ* study<sup>20</sup> (dentine discs were kept in oral cavity) showed no significant difference in the dentinal tubule occlusion score between Novamin and arginine containing toothpastes. The discrepancy in the result may be due to the different experimental condition employed in these studies.

Apart from the visual scoring index, the occlusion of dentinal tubules was also assessed based on the percentage of occluded tubules before and after tooth brushing. The results however, revealed no significant difference in the occluding capacity between Novamin and arginine containing toothpastes (Fig. 3B). The methods of assessments used in this study may have contributed to this outcome as the scoring method tends to be more specific where a score was given for the degree of dentinal tubules occlusion while in the percentage method the dentinal tubules were considered as being occluded regardless whether the tubules were partially or completely occluded.

With increasing consumption of soft drinks and acidic beverages in modern lifestyle, incidences of dental erosion are becoming more common.<sup>3, 24, 25</sup> Dental erosion is one of the major contributing factors in the initiation of dentine hypersensitivity.<sup>24, 26</sup> The occurrence of dentine hypersensitivity among the older age group is more common as there is a tendency to retain natural teeth longer due to improvements in medical and dental care.<sup>25</sup> Therefore, a sound understanding of the efficacy of the ingredients contained in toothpastes to withstand challenges of acidic beverages is important. In this study, Novamin was found to be significantly more efficient in withstanding acidic challenge of soft drinks when compared to arginine containing toothpaste and the control toothpaste. The efficacy of Novamin to withstand acidic beverage challenge was also evident in previous *in vitro* studies.<sup>8, 18-20</sup> A study showed that dentinal tubules occluded by Novamin containing toothpaste remained occluded when exposed to a constant flow of either Coca-Cola® or grapefruit juice for 5 minutes (running at 1ml/s).<sup>8</sup> It was also reported that the application of the Novamin toothpaste resulted in the formation of a new hydroxyapatite-like layer on the dentin surface and within the tubules, which provides resistance to mechanical challenges. Parkinson and Willson<sup>19</sup>, demonstrated that the dentinal tubules of bovine dentine occluded by Novamin containing toothpaste showed significantly better resistance to acid when compared to

arginine containing toothpaste following four days of twice-daily brushing where for the last two days, the dentin disc were exposed to grapefruit juice. On the other hand, Wang et al<sup>18</sup> reported that the efficacy to withstand acid (6% citric acid, pH 1.5, for 1 min) challenge was better for potassium nitrate containing toothpaste compared to that of Novamin containing toothpaste. In another study,<sup>20</sup> where treated dentine discs were kept in the oral environment (in situ model), it was found that the efficacy to withstand acid challenge between Novamin and arginine containing toothpastes was not significantly different although the score for Novamin was lower than arginine (indicating better occlusion). In that study, the discs were brushed twice daily for 10 seconds and grapefruit juice was used for acid challenge. The discrepancy of findings may be due to the difference in the methodology and the variability of the pH associate with different type of beverages.

In conclusion, the findings suggested that the occluding capacity of Novamin containing toothpaste was better than the arginine containing toothpaste. The former was also more resistant to acidic challenge of soft drinks compared to arginine containing tooth paste.

### **Conflict of Interest**

The authors declare that they have no conflict of interest.

### **References**

1. [Splieth CH, Tachou A. Epidemiology of dentin hypersensitivity. Clin Oral Investig 2013; 17 Suppl 1: S3-8.](#)
2. [Addy M. Tooth brushing, tooth wear and dentine hypersensitivity--are they associated? Int Dent J 2005; 55: 261-267.](#)

3. [Dababneh Rh, Khouri At, Addy M. Dentine hypersensitivity - an enigma? A review of terminology, mechanisms, aetiology and management. Br Dent J 1999; 187: 606-611; discussion 603.](#)
4. Shiau HJ. Dentin hypersensitivity. J Evid Based Dent Pract 2012; 12: 220-228.
5. Brannstrom M, Astrom A. The hydrodynamics of the dentine; its possible relationship to dentinal pain. Int Dent J 1972; 22: 219-227.
6. Vieira AH, Santiago SL. Management of dentinal hypersensitivity. Gen Dent 2009; 57: 120-126; quiz 127-128.
7. [Layer TM. Development of a fluoridated, daily-use toothpaste containing NovaMin technology for the treatment of dentin hypersensitivity. J Clin Dent 2011; 22: 59-61.](#)
8. [Earl JS, Leary RK, Muller KH, et al. Physical and chemical characterization of dentin surface following treatment with NovaMin technology. J Clin Dent 2011; 22: 62-67.](#)
9. [Pradeep AR, Sharma A. Comparison of clinical efficacy of a dentifrice containing calcium sodium phosphosilicate to a dentifrice containing potassium nitrate and to a placebo on dentinal hypersensitivity: a randomized clinical trial. J Periodontol 2010; 81: 1167-1173.](#)
10. [Rajesh KS, Hedge S, Arun Kumar MS, et al. Evaluation of the efficacy of a 5% calcium sodium phosphosilicate \(Novamin\) containing dentifrice for the relief of dentinal hypersensitivity: a clinical study. Indian J Dent Res 2012; 23: 363-367.](#)
11. Neuhaus KW, Milleman JL, Milleman KR, et al. Effectiveness of a calcium sodium phosphosilicate-containing prophylaxis paste in reducing dentine hypersensitivity immediately and 4 weeks after a single application: a double-blind randomized controlled trial. J Clin Periodontol 2013; 40: 349-357.
12. Carson SJ. Possible role for arginine-containing toothpastes in managing dentine hypersensitivity. Evid Based Dent 2013; 14: 44-45.

13. [Sharif MO, Iram S, Brunton PA. Effectiveness of arginine-containing toothpastes in treating dentine hypersensitivity: a systematic review. J Dent 2013; 41: 483-492.](#)
14. Que K, Fu Y, Lin L, et al. Dentin hypersensitivity reduction of a new toothpaste containing 8.0% arginine and 1450 ppm fluoride: an 8-week clinical study on Chinese adults. *Am J Dent* 2010; 23 Spec No A: 28A-35A.
15. [Talioti E, Hill R, Gillam DG. The Efficacy of Selected Desensitizing OTC Products: A Systematic Review. ISRN Dent 2014; doi: 10.1155/2014/865761.](#)
16. [Seong J, Macdonald E, Newcombe RG, et al. In situ randomised trial to investigate the occluding properties of two desensitising toothpastes on dentine after subsequent acid challenge. Clin Oral Investig 2013; 17: 195-203.](#)
17. Olley RC, Pilecki P, Hughes N, et al. An in situ study investigating dentine tubule occlusion of dentifrices following acid challenge. *J Dent* 2012; 40: 585-593.
18. [Wang Z, Sa Y, Sauro S, Chen H, et al. Effect of desensitising toothpastes on dentinal tubule occlusion: a dentine permeability measurement and SEM in vitro study. J Dent 2010; 38: 400-410.](#)
19. [Parkinson CR, Willson RJ. A comparative in vitro study investigating the occlusion and mineralization properties of commercial toothpastes in a four-day dentin disc model. J Clin Dent 2011; 22: 74-81.](#)
20. [West NX, Macdonald EL, Jones SB, et al. Randomized in situ clinical study comparing the ability of two new desensitizing toothpaste technologies to occlude patent dentin tubules. J Clin Dent 2011; 22: 82-89.](#)
21. [Prati C, Venturi L, Valdre G, et al. Dentin morphology and permeability after brushing with different toothpastes in the presence and absence of smear layer. J Periodontol 2002; 73: 183-190.](#)

22. Arrais CA, Micheloni CD, Giannini M, et al. Occluding effect of dentifrices on dentinal tubules. *J Dent* 2003; 31: 577-584.
23. West N, Addy M, Hughes J. Dentine hypersensitivity: the effects of brushing desensitizing toothpastes, their solid and liquid phases, and detergents on dentine and acrylic: studies in vitro. *J Oral Rehabil* 1998; 25: 885-895.
24. Dugmore C, Rock W. A multifactorial analysis of factors associated with dental erosion. *Br Dent J* 2004; 196: 283-286.
25. Zero DT. Etiology of dental erosion—extrinsic factors. *Eur J Oral Sci* 1996; 104: 162-177.
26. Absi EG, Addy M. Dentine hypersensitivity—the effect in vitro of acids and dietary substances on root-planned and burred dentine. *J Clin Periodontol* 1987; 14: 274-279.

**Table 1:** Major constituents of toothpastes used in this study.

| <b>Toothpastes</b>             | <b>Active ingredient(s)</b>   | <b>Fluoride content</b>                               |
|--------------------------------|---|---|
| Novamin containing toothpaste  | Calcium sodium phosphosilicate (Novamin), silicon dioxide, and glycerin | Sodium monofluorophosphate (1450 ppm F <sup>-</sup> ) |
| Arginine containing toothpaste | Arginine (8 wt%), calcium carbonate                                     | Sodium monofluorophosphate (1450 ppm F <sup>-</sup> ) |
| Control toothpaste             | -   | Sodium monofluorophosphate (1450 ppm F <sup>-</sup> ) |

## Figure Legends

**Fig. 1.** A schematic representation of the tubule occlusion level of visual scoring index used in this study. Tubules occlusion was determined using a visual scoring index of 1 to 5 (1-occluded; 2-partially unoccluded; 3-equally occluded/unoccluded; 4-partially occluded; 5-unoccluded). Note that for the score 2, 3 and 4, tubule occlusion was similar but not exactly the same to the schematic representations.

**Fig. 2.** Dentine disc with patent dentinal tubules (A). Dentine discs treated with Novamin (B), arginine (C) and control (D) toothpastes. Note that dentinal tubules were better occluded when treated with Novamin containing toothpaste compared to others.

**Fig. 3.** Dentinal tubules occlusion score (A) and percentage of occluded tubules (B) for different toothpastes. Note that dentinal tubules occluded by Novamin containing toothpaste withstand the acidic soft drink challenge comparatively for longer time period.

‘a’-denotes significant difference ( $P < 0.05$ ) at various time points when compared to before acidic soft drink challenge for respective toothpaste groups (one way ANOVA followed by Dunnett's test).

‘b’-denotes significant difference ( $P < 0.05$ ) between Novamin and arginine containing toothpastes at various time points (One way ANOVA followed by Tukey Test). ‘c’- denotes significant difference ( $P < 0.05$ ) between Novamin and control toothpastes at various time points (One way ANOVA followed by Tukey Test).

Fig. 2

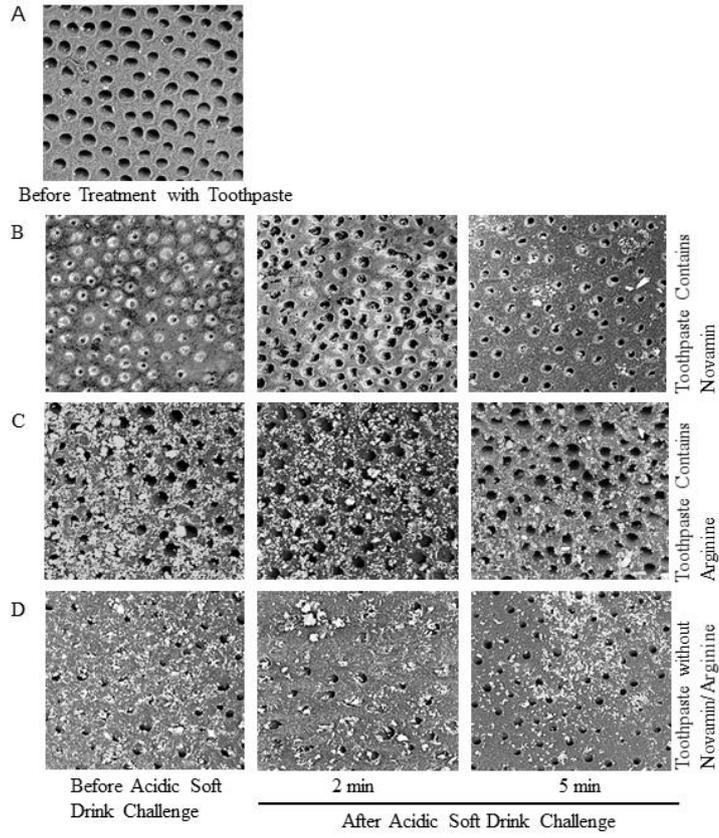


Fig. 3

